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(54) **Process for obtaining surface modified anticancer nanoparticles**

Verfahren zur Herstellung oberflächenmodifizierter Antikrebsmittel-Nanopartikel

Procédé pour obtenir des nanoparticules d'un agent anticancereux avec une surface modifiée

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EP-A- 0 262 560	EP-A- 0 499 299
WO-A-90/15593	WO-A-91/06292
WO-A-92/03380	FR-A- 2 118 987

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Description

[0001] This invention relates to a process for obtaining anticancer agents in the form anticancer compositions comprising particles, and to the use of the compositions comprising the particles.

[0002] The therapeutic index is a measure of how selective a drug is at producing its desired effects and can be defined as the ratio of the median lethal dose to the median effective dose, i.e. (LD_{50}/ED_{50}) (see Goodman and Gilman, *The Pharmacological Basis of Therapeutics*, Eighth Edition, p. 68-69). Virtually all anticancer agents have a low therapeutic index, e.g. less than about 1.0. Increasing the therapeutic index, e.g. by reducing toxicity or enhancing efficacy would provide more latitude to physicians in their duty of administering anticancer drugs to patients in need thereof. Consequently, methods to reduce toxicity and/or enhance efficacy of anticancer drugs and thus increase the therapeutic indices of such drugs would be of great value in the treatment of cancers.

[0003] In addition, poorly water-soluble drugs, such as poorly water-soluble anticancer agents, are not readily injectable via an intravenous (IV) bolus injection. The creation of injectable forms of poorly soluble drugs represents a formidable problem. It would be highly desirable to be able to provide poorly soluble drugs, such as poorly soluble anticancer agents, in an IV bolus injectable form.

[0004] The invention is as set out in claims 1 - 19.

[0005] Anticancer compositions comprising anticancer agents in the form of surface modified nanoparticles as obtained through the process according to the invention exhibit reduced toxicity and/or enhanced efficacy.

[0006] More particularly, in accordance with this invention as claimed, there is provided a process for obtaining a composition comprising anticancer particles consisting essentially of a crystalline anticancer agent having a surface modifier adsorbed on the surface thereof in an amount sufficient to maintain an effective average particle size of less than 400 nm.

[0007] In another embodiment of the invention, there is provided the use of the above-described composition as obtained through the process of the invention for the preparation of a medicament for the treatment of cancer.

[0008] It is a particularly advantageous feature of this invention that a process for obtaining anticancer compositions is provided, which compositions exhibit reduced toxicity and/or improved efficacy.

[0009] It is a further advantageous feature of this invention that the use of compositions is provided featuring poorly soluble anticancer agents that can be administered by IV bolus injection and that can exhibit prolonged circulation in the blood pool after such injection.

[0010] This invention is based partly on the discovery that surface modified anticancer nanoparticles exhibit reduced toxicity and/or enhanced efficacy.

[0011] The particles obtained with the process of this invention comprise an anticancer agent as defined in claim 1. The anticancer agent is present in one or more discrete crystalline phases. The crystalline phase differs from an amorphous, i.e. noncrystalline phase which results from conventional solvent precipitation techniques for the preparation of particles in the submicron size range, such as described in U.S. Patent No. 4,826,699.

[0012] EP 499299, which was secret at the priority date of the application covering the present invention and thus constitutes prior art with regard to the present invention only within the provision of Art. 54(3) EPC, discloses nanoparticles with dimensions similar to the ones of the present invention, where the nanoparticles of EP499299 contain drug substances other than the ones covered by the present invention for pharmaceutical purposes other than the treatment of cancer.

[0013] The nanoparticles obtained with the process according to the present invention further differ from those obtained according to EP 262560 in that the latter document discloses fine particles with an average size not larger than 1μm containing benzoyl urea compounds as the pharmaceutically active substance. EP 262 560 does not mention a screening process in order to match surfactants and active agents and ensure that the thus obtained particle maintains its desired size.

[0014] Finally, WO 90/15593 discloses a process for the preparation of drug particles consisting in the emulsification of the drug in an organic solution and the subsequent removal of the solvent, which results in drug precipitation. No mention is made in WO 90/15593 of a screening process which allows to match surfactants and active agents in order to obtain particles with optimal stability.

[0015] The invention is practised with the anticancer agents as specified in claim 1. However, the anticancer agent must be poorly soluble and dispersible in at least one liquid medium. By "poorly soluble", it is meant that the drug substance has a solubility in the liquid dispersion medium, e.g. water, of less than about 10 mg/ml, and preferably, of less than 1 mg/ml at processing temperature, e.g. room temperature. The preferred liquid dispersion medium is water. However, the invention can be practised with other liquid media in which the anticancer agent is dispersible including, for example, aqueous salt solutions, safflower oil, and solvents such as ethanol, t-butanol, hexane, and glycol. The pH of the aqueous dispersion media can be adjusted by techniques known in the art.

[0016] The anticancer agent is as defined in claim 1.

[0017] The anticancer agents useful in the practice of this invention are known compounds and/or can be prepared

by techniques known in the art.

[0018] The anticancer agent can be used alone or in combination with one or more anticancer agents.

[0019] The compositions obtained with the process of this invention contain an anticancer agent as described above having a surface modifier adsorbed on the surface thereof. Useful surface modifiers are believed to include those which physically adhere to the surface of the anticancer agent but do not chemically bond to the anticancer agent.

[0020] Suitable surface modifiers can preferably be selected from known organic and inorganic pharmaceutical excipients. Such excipients include various polymers, low molecular weight oligomers, natural products and surfactants. Preferred surface modifiers include nonionic and anionic surfactants. Representative examples of excipients include gelatin, casein, lecithin (phosphatides), gum acacia, cholesterol, tragacanth, stearic acid, benzalkonium chloride, calcium stearate, glyceryl monostearate, cetostearyl alcohol, cetomacrogol emulsifying wax, sorbitan esters, polyoxyethylene alkyl ethers, e.g., macrogol ethers such as cetomacrogol 1000, polyoxyethylene castor oil derivatives, polyoxyethylene sorbitan fatty acid esters, e.g., the commercially available Tweens™, polyethylene glycols, polyoxyethylene stearates, colloidol silicon dioxide, phosphates, sodium dodecylsultate, carboxymethylcellulose calcium, carboxymethylcellulose sodium, methylcellulose, hydroxyethylcellulose, hydroxypropylcellulose, hydroxypropylmethylcellulose phthalate, noncrystalline cellulose, magnesium aluminum silicate, triethanolamine, polyvinyl alcohol (PVA), and polyvinylpyrrolidone (PVP). Most of these excipients are described in detail in the *Handbook of Pharmaceutical Excipients*, published jointly by the American Pharmaceutical Association and The Pharmaceutical Society of Great Britain, the Pharmaceutical Press, 1986. The surface modifiers are commercially available and/or can be prepared by techniques known in the art. Two or more surface modifiers can be used in combination.

[0021] Particularly preferred surface modifiers include PVP , tyloxapol, polaxomers, such as Pluronic™ F68, F108 and F127, which are block copolymers of ethylene oxide and propylene oxide available from BASF, and poloxamines, such as Tetronic™ 908 (T908), which is a tetrafunctional block copolymer derived from sequential addition of ethylene oxide and propylene oxide to ethylenediamine available from BASF, dextran, lecithin, Aerosol OT™ (AOT), which is a dioctyl ester of sodium sulfosuccinic acid, available from American Cyanamid, Duponol™ P, which is a sodium lauryl sulfate, available from DuPont, Triton™ X-200, which is an alkyl aryl polyether sulfonate, available from Rohm and Haas, Tween 20, 40, 60 and 80, which are polyoxyethylene sorbitan fatty acid esters, available from ICI Speciality Chemicals, Span 20, 40, 60 and 80, which are sorbitan esters of fatty acids, Arlacel 20, 40, 60 and 80, which are sorbitan esters of fatty acids, available from Hercules Inc., Carbowax™ 3550 and 934, which are polyethylene glycols available from Union Carbide, Crodesta™ F-110, which is a mixture of sucrose stearate and sucrose distearate, available from Croda Inc., Crodesta SL-40, which is available from Croda Inc., hexyldecyl trimethyl ammonium chloride (CTAC), bovine serum albumin and SA90HCO, which is $C_{18}H_{37}CH_2(CON(CH_3)CH_2(CHOH)_4CH_2OH)_2$. Surface modifiers which have been found to be particularly useful include PVP, Pluronic F-108, PVA and gum acacia.

[0022] The surface modifier is adsorbed on the surface of the anticancer agent in an amount sufficient to maintain an effective average particle size of less than 400 nm. The surface modifier does not chemically react with the anticancer agent or itself. Furthermore, the individually adsorbed molecules of the surface modifier are essentially free of intermolecular crosslinkages.

[0023] As used herein, particle size refers to a number average particle size as measured by conventional particle size measuring techniques well known to those skilled in the art, such as sedimentation field flow fractionation, photon correlation spectroscopy, or disk centrifugation. By "an effective average particle size of less than 400 nm" it is meant that at least 90% of the particles have a number average particle size of less than 400 nm when measured by the above-noted techniques. In particularly preferred embodiments of the invention, the effective average particle size is less than 300 nm. with reference to the effective ,average particle size, it is preferred that at least 95% and, more preferably, at least 99% of the particles have a particle size of less than the effective average, e.g. 400 nm. In particularly preferred embodiments, essentially all of the particles have a size less than 400 nm.

[0024] Motoyama et al, U.S. Patent 4,540 602 disclose that a solid drug can be pulverized in an aqueous solution of a water-soluble high molecular substance, and that as a result of such wet grinding, the drug is formed into finely divided particles ranging from 0.5 μm or less to 5 μm in diameter. However, there is no suggestion that particles having an average particle size of less than about 1 μm can be obtained. Attempts to reproduce the wet grinding procedures described by Motoyama et al resulted in particles having an average particle size of much greater than 1 μm.

[0025] The compositions obtained with the process of this invention as defined in claim 1 can be prepared by a method comprising the steps of dispersing an anticancer agent in a liquid dispersion medium and applying mechanical means in the presence of grinding media to reduce the particle size of the anticancer agent to an effective average particle size of less than 400 nm. The particles can be reduced in size in the presence of a surface modifier, wherein the surface modifier is selected so as to be compatible with the anticancer agent through a screening process so that the dispersion exhibits no flocculation or particle agglomeration visible to the naked eye and particularly when viewed under the optical microscope at 1000x at least two days after preparation. Alternatively, the particles can be contacted with a surface modifier after attrition.

[0026] The general procedure for preparing the particles of this invention is set forth below. The anticancer agent

selected is obtained commercially and/or prepared by techniques known in the art in a conventional coarse form. It is preferred, but not essential, that the particle size of the coarse anticancer agent selected be less than about 100 μm as determined by sieve analysis. If the coarse particle size of the anticancer agent is greater than about 100 μm, then it is preferred that the particles of the anticancer agent be reduced in size to less than 100 μm using a conventional milling method such as airjet or fragmentation milling.

[0027] The coarse anticancer agent selected can then be added to a liquid medium in which it is essentially insoluble to form a premix. The concentration of the anticancer agent in the liquid medium can vary from about 0.1 - 60%, and preferably is from 5 - 30% (w/w). It is preferred, but not essential, that the surface modifier be present in the premix. The concentration of the surface modifier can vary from about 0.1 to about 90%, and preferably is 1 - 75%, more preferably 20-60%, by weight based on the total combined weight of the drug substance and surface modifier. The apparent viscosity of the premix suspension is preferably less than about 1000 centipoise.

[0028] The premix can be used directly by subjecting it to mechanical means to reduce the average particle size in the dispersion to less than 400 nm. It is preferred that the premix be used directly when a ball mill is used for attrition. Alternatively, the anticancer agent and, optionally, the surface modifier, can be dispersed in the liquid medium using suitable agitation, e.g. a roller mill or a Cowles type mixer, until a homogeneous dispersion is observed in which there are no large agglomerates visible to the naked eye. It is preferred that the premix be subjected to such a premilling dispersion step when a recirculating media mill is used for attrition.

[0029] The mechanical means applied to reduce the particle size of the anticancer agent conveniently can take the form of a dispersion mill. Suitable dispersion mills include a ball mill, an attritor mill, a vibratory mill, a planetary mill, media mills such as a sand mill and a bead mill. A media mill is preferred due to the relatively shorter milling time required to provide the intended result, i.e. the desired reduction in particle size. For media milling, the apparent viscosity of the premix preferably is from about 100 to about 1000 centipoise. For ball milling, the apparent viscosity of the premix preferably is from about 1 up to about 100 centipoise. Such ranges tend to afford an optimal balance between efficient particle fragmentation and media erosion.

[0030] The grinding media for the particle size reduction step can be selected from rigid media preferably spherical or particulate in form having an average size less than about 3 mm and, more preferably, less than about 1 mm. Such media desirably can provide the particles of the invention with shorter processing times and impart less wear to the milling equipment. The selection of material for the grinding media is not believed to be critical. However, zirconium oxide, such as 95% ZrO stabilized with magnesia, zirconium silicate, and glass grinding media provide particles having levels of contamination which are believed to be acceptable for the preparation of pharmaceutical compositions. Further, other media, such as stainless steel, titania, alumina, and 95% ZrO stabilized with yttrium, are expected to be useful. Preferred media have a density greater than about 2.5 g/cm^3.

[0031] The attrition time can vary widely and depends primarily upon the particular mechanical means and processing conditions selected. For ball mills, processing times of up to five days or longer may be required. On the other hand, processing times of less than 1 day (residence times of one minute up to several hours) have provided the desired results using a high shear media mill.

[0032] The particles must be reduced in size at a temperature which does not significantly degrade the anticancer agent. Processing temperatures of less than about 30 - 40°C are ordinarily preferred. If desired, the processing equipment can be cooled with conventional cooling equipment. The method is conveniently carried out under conditions of ambient temperature and at processing pressures which are safe and effective for the milling process. For example, ambient processing pressures are typical of ball mills, attritor mills and vibratory mills. Processing pressures up to about 20 psi (1.4 kg/cm^2) are typical of media milling.

[0033] The surface modifier, if it was not present in the premix, must be added to the dispersion after attrition in an amount as described for the premix above. Thereafter, the dispersion can be mixed, e.g. by shaking vigorously. Optionally, the dispersion can be subjected to a sonication step, e.g. using an ultrasonic power supply. For example, the dispersion can be subjected to ultrasonic energy having a frequency of 20 - 80 kHz for a time of about 1 to 120 seconds.

[0034] The relative amount of the anticancer agent and surface modifier can vary widely and the optimal amount of the surface modifier can depend, for example, upon the particular anticancer agent and surface modifier selected, the critical micelle concentration of the surface modifier if it forms micelles, the surface area of the anticancer agent, etc. The surface modifier preferably is present in an amount of about 0.1-10 mg per square meter surface area of the anticancer agent. The surface modifier can be present in an amount of 0.1-90%, preferably 0.5-80%, and more preferably 1-60% by weight based on the total weight of the dry particle.

[0035] A simple screening process has been developed whereby compatible surface modifiers and anticancer agents can be selected which provide stable dispersions of the desired particles. First, coarse particles of an anticancer agent are dispersed in a liquid in which the anticancer agent is essentially insoluble, e.g. water at 2% (w/v) and milled for 120 hours in a roller mill under the following milling conditions:

Grinding vessel:	8 oz. (250 ml) glass jar
Available volume of grinding vessel:	250 ml
Media volume:	120 ml
Media type:	1.0 mm pre-cleaned zirconium oxide beads (distributed by Zircoa Inc.)
Milling time:	120 hours
Slurry volume:	60 ml
RPM:	92
Room Temperature	

[0036] The slurry is separated from the milling media by conventional means, e.g. by pouring the slurry out of the vessel, or by using a pipette. The separated slurry is then divided into aliquots and surface modifiers are added at a concentration of between 2 and 50% by weight based on the total combined weight of the anticancer agent and surface modifier. The dispersions are then sonicated (1 minute, 20 kHz) or vortexed using a multitubed vortexer for one minute, to disperse agglomerates and subjected to particle size analysis, e.g. by photon correlation spectroscopy (PCS) and/or by examination under an optical microscope (1000 x magnification). If a stable dispersion is observed, then the process for preparing the particular anticancer agent/surface modifier combination can be optimized in accordance with the teachings above. By stable it is meant that the dispersion exhibits no flocculation or particle agglomeration visible to the naked eye and, preferably, when viewed under the optical microscope at 1000x at least two days or longer, after preparation. In addition, preferred particles exhibit no flocculation or agglomeration when dispersed in 0.1 N HC1 and/or phosphate buffered saline, pH 7.4 (PBS) or rat plasma.

[0037] The resulting dispersion is stable and consists of the liquid dispersion medium and the above-described particles. The dispersion of surface modified anticancer agent nanoparticles can be spray coated onto sugar spheres or onto a pharmaceutical excipient in a fluid-bed spray coater by techniques well known in the art.

[0038] Anticancer pharmaceutical compositions obtained with the process according to this invention include the particles described above and a pharmaceutically acceptable carrier therefor. Suitable pharmaceutically acceptable carriers are well known to those skilled in the art. These include non-toxic physiologically acceptable carriers, adjuvants or vehicles for parenteral injection, for oral administration in solid or liquid form, for rectal administration, nasal administration, intramuscular administration, subcutaneous administration, and the like.

[0039] The use of the composition as obtained by means of the process described herein for preparing a medicament for the treatment of cancer is such that an effective amount of the above-described anticancer composition can then be administered to a mammal in need of treatment. The selected dosage level of the anticancer agent for treatment is effective to obtain a desired therapeutic response for a particular composition and method of administration. The selected dosage can be readily determined by one skilled in the art and depends upon the particular anticancer agent, the desired therapeutic effect, the route of administration, the desired duration of treatment and other factors.

[0040] It is a particularly advantageous feature that the anticancer compositions obtained with the method of this invention exhibit reduced toxicity and/or enhanced efficacy as illustrated in the examples that follow. Further, the particles obtained with the method of this invention exhibit prolonged circulation in the blood pool.

[0041] Moreover, anticancer agents which heretofore could not be administered by injection, when prepared as nanoparticles and formulated in anticancer compositions according to the process of this invention, can be effectively administered by injection, e.g. by an IV bolus injection.

[0042] The following examples 1-13 illustrate the invention, which is in no way, however, to be construed as limited thereto.

<u>Example 1 - 6 Nanoparticulate Taxol</u>

<u>Example 1</u>

[0043] Approximately 18 ml precleaned zirconium oxide media (1 mm) was added to a 30 ml amber jar. To it was added 240 mg taxol (Sigma Chemicals) and 180 mg Tween 20. Finally, 12 ml water for injection was added to the jar, it was sealed and mounted on a roller mill for 11 days. The final particle size was 327 nm. The formulation was stable when exposed to PBS (pH 7.4) and rat plasma.

Example 2

[0044] Example 1 was repeated except that the Tween 20 was replaced with PVA (MW 30 to 70 k). The final particle size was 365 nm.

[0045] The above samples were evaluated in efficacy studies in mice bearing early stage mammary adenocarcinoma #16/C. The antitumor activity was assessed by comparing tumor weights of taxol treated animals with the tumor weights of untreated animals. The toxicity was assessed by dose ranging studies with death and weight loss as end points. All samples were injected IV.

Example	Dose mg/kg	%Wt Loss	Deaths	Median Tumor on Day 11	T/C %
Control D	----	+ 6.1	----	1539 mg	--
Example 1	108.5	- 1.5	0/5	1528	13
Example 2	108.5	- 3.0	0/5	201	99

[0046] A control sample of taxol (NCI) was not available. However, a single dose of taxol formulated in Cremophore EL causes immediate deaths at 25 mg/kg total dose. However, taxol formulated in compositions of this invention could be injected at a dose of 88 mg/kg with no apparent adverse effects.

[0047] A taxol suspension prepared in a manner similar to Example 1 was treated separately with several surface modifiers. After addition of the new surface modifier the mixture was vortexed and evaluated for particle size and fluid stability. All the suspensions contained 1% taxol and 0.75% Tween 20. The results are as follows.

Example/ 3-6

[0048]

Surface Modifier	Concentration %	Size(nm)	Fluid Stability	
			PBS	Rat Plasma
3 CTAC	0.25	364	OK	Flocculation
4 AOT	0.25	322	SA*	SA
5 F68	0.5	297	SA/OK	SA/OK
6 T908	0.5	313	SA/OK	SA/OK

*SA = Slight Aggregation

Examples 7- 8 Nanoparticulate 1,2,4-benrotriazin-7-amine 1,4-dioxide

[0049] Approximately 60 ml precleaned zirconium oxide (1 mm) media was transferred into a 4 oz amber jar. It was followed by addition of 1.5 g 1,2,4-benzotriazin-7-amine 1,4-dioxide and 28.5 ml water for injection. The jars were sealed, loaded onto a roller mill and cascaded at 95 RPM for 48 hrs. PCS analysis determined the particle size to be 322 nm, however, the presence of larger particles was suggested. Milling was continued for 5 additional days.

[0050] Studies were conducted by mixing 0.5 ml of the benzotriazinamine dioxide slurry prepared above with 0.5 ml 6% surface modifier solutions. The final concentration of the drug was 2.5% and that of the surface modifiers was 3%. The surface modifier stabilized nanosuspensions of benzotriazinamine dioxide were then treated with either PBS (pH 7.0) or 0.1 N HCl (pH 1). Optical microscopic observations were made to determine fluid stability. The results are as follows;

Example	Surface Modifier	Stability		Human Plasma
		pH 1	pH 7	
7	PVP (12K) (BASF)	Fine	Fine	Fine
8	Gum Acacia (Aldrich)	----	SA/OK	SA/OK

[0051] It was concluded that stable nanoparticles of 1,2,4-benzo-triazin-7-amine 1,4-dioxide could be prepared.

Examples 9-12 Nanoparticulate 1,2,4-benzotriazin-3-amine 1,4-dioxide

[0052] 7.5 ml precleaned zirconium oxide media (1 mm) was transferred into a 15 ml amber jar along with 18.75 mg 1,2,4-benzotriazin-3-amine 1,4-dioxide and 3.75 ml water. After 11 days of milling the nanosuspension was separated from the media. To each of the 100 μl aliquots of the suspension, 100 μl of a surfactant solution (2%) was added giving a final concentration of 0.25% drug and 1% surfactant. The mixture was vortexed and analyzed for particle size. Fluid stability was assessed microscopically by mixing 10 μl of the suspension with 90 μl of rat plasma. The results are as follows:

Example	Surface Modifier	Particle Size(nm)	Fluid Stability Rat Plasma
9	PVP (12K)	134	Fine
10	Gum Acacia	344	SA/OK
11	Tween 80	128	Fine
12	T908	130	Aggregates

Example 13 Nanoparticulate Retinoic Acid

[0053] 30 ml precleaned zirconium oxide media was transferred to a 60 ml amber jar. To it was added 1 g transretinoic acid (Sigma), 470 mg tyloxapol and 15 ml water. The mixture was milled on a roller mill for 15 days. The final particle size was 140 nm. The nanosuspension was stable when exposed to either rat plasma or simulated gastric fluid.

[0054] Examples 14-23 do not refer to the subject-matter claimed but are useful for illustrating synthetic methods applicable to preparing compositions according to the process of the invention. The compounds used in the following examples 14-23 are disclosed in EP 499 299 and do not form part of the present invention.

Examples 14-17 Nanoparticulate Piposulfan Example 14

[0055] Piposulfan (purchased from Eastman Kodak) was milled in a mixture of 0.33% polyoxyethylene sorbitan monooleate, Tween 80, (ICI Americas Inc., Wilmington DE) and 0.67% sorbitan monooleate, Span 80, (ICI) using 1 mm zirconium oxide beads for about 96 hours to produce particles approximately 240 nm in diameter. The final piposulfan concentration in the suspension was 10 mg/ml. The particles were stable to flocculation/aggregation in rat plasma.

[0056] Milling Conditions: A coarse suspension of piposulfan was prepared by adding 300 mg of the drug to a 4 oz. (120 ml) amber bottle which was previously filled with 60 ml of 1 mm precleaned zirconium oxide beads (Zircoa Inc., Solon OH) and 30 ml 1% Tween 80/-Span 80 (1 to 2 ratio) solution. The surfactant solution was prepared by accurately weighing 333 mg Tween 80 and 667 mg Span 80 in a volumetric flask followed by addition of sterile water for injection to dissolve/disperse the surfactants. Sufficient quantity of water was added to make the final volume 100 ml. Zirconium oxide beads were cleaned by first rinsing in 1N sulfuric acid followed by several rinses with deionized water. The media was dried in a vacuum oven at about 100°C overnight.

[0057] The sealed primary container was loaded into a secondary padded aluminium containment can to ensure a tight fit. It was milled on a roller mill (US Stoneware, Mawah, NJ) at 144 RPM for about 96 hours. At the end of the milling time the slurry was separated from the media and particle size was measured using a PCS device. Stability of these

particles to rat plasma was assessed by optical microscopy at 1000 X magnification. The final pH of the formulation was 6.

[0058] Control A (unmilled) A coarse suspension containing 40 mg bulk piposulfan was dispersed in water in the presence of 3% ethanol and 1% Tween 80. This suspension could not be injected IV.

[0059] Example 14 was evaluated for efficacy studies in female mice (average weight 22 g) which were implanted with early stage Mammary Adenocarcinoma # 16/C on day 0. The formulation was injected starting from day 1 for several days. The antitumor activity was assessed by monitoring tumor weight and comparing it to the control animals. The results were as follows:

Treatment	Route of Adm.*	Total Dose (mg/kg)	% Wt. Loss	T/C %	Log10 Cell Kill
Control	----	---	+5.5	---	----
Example 14	IV	356	-5.5	0	2.75
(243 nm)	IV	220	-5.5	2	2.75
	IV	137	-1.8	2	2.25
	IV	85	0	18	1.0
Control A	SC	800	-10.8	0	2.1

* Administration - IV= Intravenous; SC= Subcutaneous

Example 1 could be injected directly as a 10 mg/ml suspension. There was no acute toxicity after injection of 78 mg/kg single dose.

$$T/C = \frac{\text{Tumor weight in treated animals}}{\text{Tumor weight of control animals}} \text{ as \% value.}$$

[0060] Lower value indicates better efficacy, 0% suggests cures, <10% is considered highly active, 10 to 42% is a moderately active formulation, >42% is considered inactive.

$$\text{Log Kill} = (T\text{-}C)/3.32\,(Td), \text{ where}$$

T is the time in days for the median tumor to reach 1000 mg mass in treated animals,
C is the time in days for the median tumor to reach 1000 mg in control animals and
Td is the tumor volume doubling time in days.

Cures (tumor free animals) are excluded from (T-C) calculations.

[0061] The composition of Example 14 exhibited reduced toxicity and enhanced efficacy compared to other prior art compositions and could be administered by IV bolus injection.

Examples 15-17

[0062] The milling procedure described in Example 14 was repeated except that the ratio of Tween 80 to Span 80 was 2:1. The resulting average particle size was 297 nm.

[0063] The milling procedure described in Example 14 was repeated except that the ratio of Tween 80 to Span 80 was 1:1. The resulting average particle size was 380 nm.

[0064] The milling procedure described in Example 14 was repeated except that the surface modifier was a 1:1 ratio of Tween 60 and Span 60. The resulting average particle size was 301 nm.

[0065] Stable piposulfan nanoparticles were also prepared using bovine serum albumin as the surface modifier.

Examples 18-20 Nanoparticulate Camptothecin Example 18

[0066] Approximately 60 ml precleaned zirconium oxide beads (1 mm) were placed in a 120 ml wide mouth round amber bottle. To it was added 0.35 g of Tetronic 908 (BASF) followed by 0.35 g Camptothecin (Sigma Chemicals, 95% pure). To the above mixture, 35 ml water for injection (Abbott) was added. The bottle was sealed and mounted on a roller mill. Milling was effected by rotating the bottle at 100 RPM for 7 days.

[0067] At the end of milling, an aliquot (100 µl) was checked for particle size using a Malvern Zetasizer. The particles were determined to have an average particle size of 240 nm.

Example 19

[0068] Example 18 was repeated except that the Tetronic 908 was replaced by PVA (MW 30 to 70 K). The final particle size was 256 nm.

Example 20

[0069] Example 18 was repeated except that Tetronic 908 was replaced by gum acacia. The final particle size was 298 nm.

[0070] Nanocamptothecin formulations were evaluated for efficacy in two murine tumor models, i.e., Mammary Adenocarcinoma #16/C and Pancreatic Ductal Adenocarcinoma #03. The antitumor activity was assessed by monitoring tumor weight from experimental and control animals.

1. Efficacy Studies in Pancreatic Ductal Adenocarinoma #03:

Example	Route	Dose mg/kg	Wt% Loss	Drug Deaths	T/C %
Control B	SC	60	-24.1	6/6	-
	SC	40.2	-21.8	5/5	-
	SC	26.7	-18.2	5/5	-
	SC	18	-10.9	1/5	62
19	IV	83.1	-16.7	1/4	14
	IV	78.2	-14.6	1/4	55
	IV	48.6	- 8.3	0/4	0
	IV	24.3	- 4.2	0/4	18
PVA Control	IV	----	+ 6.3	0/4	100
20	IV	93.5	-16.7	1/4	7
	IV	46.8	-14.6	0/4	17
	IV	23.4	- 8.3	0/4	11
Gum Acacia Control	IV	----	0.0	0/4	60

The Control B formulation consisted of 1% coarse camptothecin in 3% ethanol, and 1% Tween 20. Control B could only be administered subcutaneously and even at the lowest SC dose (18 mg/kg) was inactive. Control B was toxic to 1/5 animals tested. In contrast, doses of the nanocamptothecin formulations prepared with the process of this invention ranging from 24-93 mg/kg were administered IV and were shown to be safe and efficacious.

2. Efficacy Studies in Mammary Adenocarcinoma #16/C Murine Tumor Model					
Example	Route	Dose mg/kg	%Wt Loss	Drug Deaths	T/C %
Control B	SC	60	-23.5	5/5	-
	SC	30	-20.9	5/5	-
	SC	15	-18.3	3/5	14**
18	IV	65	-17.4	0/5	23
	IV	33	-18.7	1/5	33
	IV	16	- 2.2	0/5	63
T908 Control	IV	--	+ 4.3	0/2	100
19	IV	65	-21.7	5/5	-
	IV	33	-15.7	0/5	100
PVA Control	IV	--	+ 4.3	0/2	100
The T908 and PVA controls consisted of 1% aqueous solutions of the respective surface modifiers. The Control B was injected subcutaneously, and it was toxic at all doses tested. Efficacious doses of the nanocamptothecin formulations prepared with the process of this invention were administered intravenously.					

** % T/C for the control animals was determined from the number(N) of the animals surviving in the sample. In this particular case N=2.

3. Blood and Tumor Clearance

[0071] To determine if the increased efficacy was related to alterations in pharmacokinetic properties, blood clearance and tumor distribution were studied in the Mammary adenocarcinoma #16/C murine tumor model.

[0072] Tumor bearing mice were injected via the tail vein with 10 mg/ml camptothecin, formulated as described in Examples 18 and 19, and a control which was 5 mg/ml camptothecin solubilized by the addition of 0.1 N NaOH. At various times after injection, i.e. 5 min., 30 min., 60 min., 2 hrs., 4 hrs., 8 hrs., 16 hrs., 24 hrs. and 48 hrs., animals were euthanized and a blood sample was collected and the tumor excised. Concentrations of drug samples were quantified using HPLC. Results show that the compositions of this invention affect the clearance of the drug from the circulating pool of blood and the tumor.

T1/2 Blood and Tumor		
Formulation	Blood	Tumor
Example 19	18 hrs.	> 48 hrs.
Example 18	13 hrs.	> 48 hrs.
Control	1.6 hrs.	13.5 hrs.

[0073] When formulated in accordance with the process of this invention, the elimination half-life and the residence time of camptothecin in the tumor were prolonged significantly. It was concluded that pharmacokinetic parameters of the nanoparticulate formulation of camptothecin as prepared with the process of the present invention are directly related to the improved performance of the drug.

21-23 Nanoparticulate Etoposide

Example 21

[0074] Example 18 was repeated except that 1.7 g etoposide was combined with 1.7 g PVA and the milling time was 14 days. The final particle size was 310 nm. The particles were stable in acid and plasma.

Example 22

[0075] Example 21 was repeated except that the PVA was replaced by Pluronic F-108 (BASF). The final particle size was 312 nm. The particles were stable in acid and plasma.

Example 23

[0076] Etoposide (2%) was milled in sterile water for 7 days. A 1:1 mixture of the milled slurry was prepared with 2% Pluronic F127 solution. The mixture was vortexed before measuring particle size. The final size was 277 nm. The slurry was stable in simulated gastric fluid, PBS (pH 7.4) and rat plasma.

Efficacy Studies

[0077] Nanoetoposide formulations were evaluated in two separate efficacy studies in pancreatic ductal adenocarcinoma #03 (PANC #03). Control C was a 2% non-aqueous etoposide solution prepared using the formula described on pages 741-743 of the 46th Edition of the Physicians' Desk Reference. As described above, antitumor activity was assessed by monitoring tumor weight from experimental and control animals. These studies demonstrate that the etoposide compositions when prepared according to the process of this invention provide a means to deliver high doses of the drug without evidence of severe toxic reaction.

1. Efficacy Studies for Nanoetoposide in PANC #03 Murine Tumor Model

Example	Route	Dose mg/kg	%Wt Loss	Drug Deaths	T/C %
Control C	IV	120	-24.0	0/5	4.0
	IV	75	- 4.0	0/5	20.0
20	IV	160	-12.0	0/5	18.0
	IV	100	0.0	0/5	32.0
	IV	62	2.0	0/5	42.0
21	IV	160	-12.0	0/5	26.0
	IV	100	+ 2.0	0/5	35.0
	IV	62	+ 4.0	0/5	35.0
22	IV	170	-18.5	1/5	16
	IV	85	- 2.0	0/5	35
	IV	43	+ 2.5	0/5	41

Claims

1. A process for obtaining a composition comprising nanoparticles consisting essentially of:

(a) a crystalline anticancer agent selected from the group consisting of chlormethine, chlorambucile, uramustine, mannomustine, mechlorethaninoxide, cyclophosphamide, ifosfamide, trifosfamide, tretamine, thiotepa, triazoquone, mitomycine, busulfan, carmustine, lomustine, semustine, streptozotocine, alkylating agents of the mitobronitole, decarbazine and procarbazine type, fluorouracil, floxuridine, tegafur, cytarabine, idoxuridine, flucytosine, mercaptopurine, azathioprine, tiamiprine, vidarabine, pentostatin, puromycine, vinblastine, vincris-

tine, teniposide, adriamycine, daunomycine, doctomycin, mithramycin, bleomycin, mitomycin, L-asparaginase, alpha-interferon, taxol, retinoic acid, prednisone, hydroxyprogesterone caproate, medroxyprogesterone acetate, megesterol acetate, diethylstilbestrol, testosterone propionate, fluoxomesterone, flutamide, leuprolide, 1,2,4-benzotriazin-3-amine 1,4-dioxide, 1,2,4-benzotriazine-7-amine 1,4-dioxide, cisplatin, carboplatin, hydroxyurea, mitotane, aminoglutethimide, cyclosporine, azathioprine, sulfasalazione, methoxsalen and thalidomide;

(b) a surface modifier adsorbed on the surface of the anticancer agent, in an amount sufficient to maintain an average particle size of less than 400 nm;

wherein the nanoparticles are mechanically obtained in a liquid dispersion medium in which the anticancer agent has a solubility of less than 10 mg/ml;

at least 90% of the particles having an average particle size of less than 400 nm;

and wherein the surface modifier is selected so as to be compatible with the anticancer agent through a screening process so that the dispersion exhibits no flocculation or particle agglomeration visible to the naked eye and particularly when viewed under the optical microscope at 1000x at least two days after preparation.

2. The process of claim 1, wherein the surface modifier is present in an amount of 0.1 to 90% by weight of the particle.

3. The process of claim 1, wherein the nanoparticles have an average particle size of less than 300 nm.

4. The process according to any one of the preceding claims, wherein the surface modifier is selected from the group comprising polyvinyl alcohol, a tetrafunctional block copolymer derived from sequential addition of ethylene oxide and propylene oxide to ethylenediamine, gum acacia, a block copolymer of ethylene oxide and propylene oxide, a polyoxyethylene sorbitan fatty acid ester and a sorbitan ester of a fatty acid.

5. The process according to any one of the preceding claims, wherein the anticancer agent is taxol.

6. The process according to any one of claims 1 to 4, wherein the anticancer agent is selected from the group consisting of 1,2,4-benzotriazin-3-amine 1,4-dioxide, 1,2,4-benzotriazine-7-amine 1,4-dioxide, and retinoic acid.

7. The process according to any one of claims 1 to 3, wherein the anticancer agent is 1,2,4-benzotriazin-3-amine 1,4-dioxide, and the surface modifier is polyvinylpyrrolidone.

8. The process according to any one of claims 1 to 3, wherein the anticancer agent is 1,2,4-benzotriazine-7-amine 1,4-dioxide and the surface modifier is gum acacia.

9. The process according to any one of claims 1 to 3, wherein the anticancer agent is 1,2,4-benzotriazine-7-amine 1,4-dioxide and said surface modifier is polyvinylpyrrolidone.

10. The process according to any one of claims 1 to 3, wherein the anticancer agent is 1,2,4-benzotriazine-7-amine 1,4-dioxide and said surface modifier is a tetrafunctional block copolymer derived from sequential addition of ethylene oxide and propylene oxide to ethylenediamine

11. The process according to any one of claims 1 to 3, wherein the anticancer agent is retinoic acid and the surface modifier is tyloxapol.

12. The process according to any one of the preceding claims, wherein the composition is injectable.

13. A process according to any one of the preceding claims wherein the composition contains a pharmaceutical carrier therefor.

14. Use of a composition as prepared with the process of any of the claims 1 to 13 for preparing a medicament for the treatment of cancer.

15. Use according to claim 14, wherein the surface modifier is present in an amount of 0.1 to 90% by weight of the particle.

16. Use according to claim 14, wherein the nanoparticles have an average particle size of less than 300 nm.

17. Use according to any one of claims 14 to 16, wherein the surface modifier is selected from the group comprising polyvinyl alcohol, a tetrafunctional block copolymer derived from sequential addition of ethylene oxide and propylene oxide to ethylenediamine, gum acacia, a block copolymer of ethylene oxide and propylene oxide, a polyoxyethylene sorbitan fatty acid ester and a sorbitan ester of a fatty acid.

18. Use according to any one of claims 14 to 17, wherein the anticancer agent is taxol.

19. Use according to any one of claims 14 to 18, wherein the composition is injectable.

Patentansprüche

1. Verfahren zur Herstellung einer Zusammensetzung, die Nanopartikel umfaßt, die im wesentlichen aus folgendem bestehen:

   (a) einem kristallinen Antikrebsmittel, gewählt aus der Gruppe bestehend aus Chlormethin, Chlorambucil, Uramustin, Mannomustin, Mechlorethaninoxid, Cyclophosphamid, Ifosfamid, Trifosfamid, Tretamin, Thiotepa, Triazochon, Mitomycin, Busulfan, Carmustin, Lomustin, Semustin, Streptozotocin, alkylierenden Mitteln des Mitobronitol-, Decarbazin- und Procarbazintyps, Fluoruracil, Floxuridin, Tegafur, Cytarabin, Idoxuridin, Flucytosin, Mercaptopurin, Azathioprin, Tiamiprin, Vidarabin, Pentostatin, Puromycin, Vinblastin, Vincristin, Teniposid, Adriamycin, Daunomycin, Doctomycin, Mithramycin, Bleomycin, Mitomycin, L-Asparaginase, alpha-Interferon, Taxol, retinoischer Säure, Prednison, Hydroxyprogesteroncaproat, Medroxyprogesteronacetat, Megesterolacetat, Diethylstilbestrol, Testosteronpropionat, Fluoxomesteron, Flutamid, Leuprolid, 1,2,4-Benzotriazin-3-amin-1,4-dioxid, 1,2,4-Benzotriazin-7-amin-1,4-dioxid, Cisplatin, Carboplatin, Hydroxyharnstoff, Mitotan, Aminoglutethimid, Cyclosporin, Azathioprin, Sulfasalazion, Methoxsalen und Thalidomid;
   (b) einem Oberflächenwandler, der auf der Oberfläche des Antikrebsmittels adsorbiert ist, in einer Menge, die ausreichend ist, um eine mittlere Partikelgröße von weniger als 400nm zu erhalten;
   wobei die Nanopartikel mechanisch in einem flüssigen Dispersionsmedium erhalten werden, in dem das Antikrebsmittel eine Löslichkeit von weniger als 10mg/ml aufweist;
   wobei mindestens 90% der Partikel eine mittlere Partikelgröße von weniger als 400nm haben;
   und wobei der Oberflächenwandler mittels eines Screeningverfahrens so ausgewählt ist, daß er mit dem Antikrebsmittel kompatibel ist, so daß die Dispersion keine Ausflockung oder Partikelagglomeration zeigt, die mit bloßem Auge und insbesondere, wenn unter dem optischen Mikroskop bei 1000x mindestens zwei Tage nach der Herstellung beobachtet, sichtbar ist.

2. Verfahren nach Anspruch 1, wobei der Oberflächenwandler in einer Menge von 0,1 bis 90 Gewichtsprozent der Partikel vorhanden ist.

3. Verfahren nach Anspruch 1, wobei die Nanopartikel eine mittlere Partikelgröße von weniger als 300nm besitzen.

4. Verfahren nach einem beliebigen der vorhergehenden Ansprüche, wobei der Oberflächenwandler aus der Gruppe gewählt ist, die Polyvinylalkohol, ein tetrafunktionelles Blockcopolymer, abgeleitet durch sequenzielle Addition von Ethylenoxid und Propylenoxid an Ethylendiamin, Gummiacacia, ein Blockcopolymer aus Ethylenoxid und Propylenoxid, einen Polyoxyethylensorbitanfettsäureester und einen Sorbitanester einer Fettsäure umfaßt.

5. Verfahren gemäß irgendeinem der vorhergehenden Ansprüche, wobei das Antikrebsmittel Taxol ist.

6. Verfahren gemäß irgendeinem der Ansprüche 1 bis 4, wobei das Antikrebsmittel aus der Gruppe gewählt ist, die aus 1,2,4-Benzotriazin-3-amin-1,4-dioxid, 1,2,4-Benzotriazin-7-amin-1,4-dioxid and retinoischer Säure besteht.

7. Verfahren gemäß irgendeinem der Ansprüche 1 bis 3, wobei das Antikrebsmittel 1,2,4-Benzotriazin-3-amin-1,4-dioxid ist, und wobei der Oberflächenwandler Polyvinylpyrrolidon ist.

8. Verfahren gemäß irgendeinem der Ansprüche 1 bis 3, wobei das Antikrebsmittel 1,2,4-Benzotriazin-7-amin-1,4-dioxid ist, und wobei der Oberflächenwandler Gummiacacia ist.

9. Verfahren gemäß irgendeinem der Ansprüche 1 bis 3, wobei das Antikrebsmittel 1,2,4-Benzotriazin-7-amin-1,4-dioxid ist, und wobei der Oberflächenwandler Polyvinylpyrrolidon ist.

10. Verfahren gemäß irgendeinem der Ansprüche 1 bis 3, wobei das Antikrebsmittel 1,2,4-Benzotriazin-7-amin-1,4-dioxid ist, und wobei der Oberflächenwandler ein tetrafunktionelles Blockcopolymer ist, das durch sequenzielle Addition von Ethylenoxid und Propylenoxid an Ethylendiamin abgeleitet ist.

11. Verfahren gemäß irgendeinem der Ansprüche 1 bis 3, wobei das Antikrebsmittel retinoische Säure ist und der Oberflächenwandler Tyloxapol ist.

12. Verfahren gemäß irgendeinem der vorherigen Ansprüche, wobei die Zusammensetzung injizierbar ist.

13. Verfahren gemäß irgendeinem der vorherigen Ansprüche, wobei die Zusammensetzung einen pharmazeutischen Träger dafür enthält.

14. Verwendung einer Zusammensetzung, wie hergestellt nach dem Verfahren gemäß irgendeinem der Ansprüche 1 bis 13 zur Herstellung eines Medikaments zur Behandlung von Krebs.

15. Verwendung gemäß Anspruch 14, wobei der Oberflächenwandler in einer Menge von 0,1 bis 90 Gewichtsprozent der Partikel vorhanden ist.

16. Verwendung gemäß Anspruch 14, wobei die Nanopartikel eine mittlere Partikelgröße von weniger als 300nm haben.

17. Verwendung gemäß irgendeinem der Ansprüche 14 bis 16, wobei der Oberflächenwandler aus der Gruppe gewählt ist, die Polyvinylalkohol, ein tetrafunktionelles Blockcopolymer, abgeleitet durch sequenzielle Addition von Ethylenoxid und Propylenoxid an Ethylendiamin, Gummiacacia, ein Blockcopolymer aus Ethylenoxid und Propylenoxid, einen Polyoxyethylensorbitanfettsäureester und einen Sorbitanester einer Fettsäure umfaßt.

18. Verwendung gemäß irgendeinem der Ansprüche 14 bis 17, wobei das Antikrebsmittel Taxol ist.

19. Die Verwendung gemäß einem der Ansprüche 14 bis 18, wobei die Zusammensetzung injizierbar ist.

Revendications

1. Procédé d'obtention d'une composition comprenant des nanoparticules essentiellement constituées de :

   (a) un agent anticancéreux cristallin choisi dans l'ensemble constitué par la chlorméthine, le chlorambucil, l'uramustine, la mannomustine, l'oxyde de méchloréthanine, le cyclophosphamide, l'ifosfamide, le trifosfamide, la tétramine, la thiotépa, la triazaquone, la mitomycine, le busulfan, la carmustine, la lomustine, la sémustine, la streptozotocine, des agents alkylants de type mitobronitole, décarbazine et procarbazine, le fluoro-uracile, la floxuridine, le tégafur, la cytarabine, l'idoxuridine, la flucytosine, la mercaptopurine, l'azathioprine, la tiamiprine, la vidarabine, la pentostatine, la puromycine, la vinblastine, la vincristine, le téniposide, l'adriamycine, la daunomycine, la doctomycine, la mithramycine, la bléomycine, la mitomycine, la L-asparaginase, l'intenféron alpha, le taxol, l'acide rétinoïque, la prednisone, le caproate d'hydroxyprogestérone, l'acétate de médroxyprogestérone, l'acétate de mégestérol, le diéthylstilbestrol, le propionate de testostérone, la fluoxomestérone, le flutamide, le leuprolide, le 1,4-dioxyde de 1,2,4-benzotriazine-3-amine, le 1,4-dioxyde de 1,2,4-benzotriazine-7-amine, le cisplatine, le carboplatine, l'hydroxyurée, le mitotane, l'aminoglutéthimide, la cyclosporine, l'azathioprine, la sulfasalazione, le méthoxsalen et le thalidomide ;
   (b) un modificateur de surface adsorbé sur la surface de l'agent anticancéreux en une quantité suffisante pour maintenir une dimension moyenne de particules inférieure à 400 nm ;
   dans lequel les nanoparticules sont obtenues mécaniquement dans un milieu liquide de dispersion dans lequel l'agent anticancéreux a une solubilité inférieure à 10 mg/ml ;
   au moins 90 % des particules ayant une dimension moyenne inférieures à 400 nm ;
   et dans lequel on choisit le modificateur de surface de façon à ce qu'il soit compatible avec l'agent anticancéreux au moyen d'un procédé de dépistage de façon à ce que la dispersion ne présente pas de floculation ou d'agglomération des particules qui soit visible à l'oeil nu et en particulier lorsqu'on l'observe au microscope optique à un grossissement de 1000 fois au moins deux jours après la préparation.

2. Procédé selon la revendication 1, dans lequel le modificateur de surface est présent en une quantité de 0,1 à 90 % en poids par rapport aux particules.

3. Procédé selon la revendication 1, dans lequel les nanoparticules ont une dimension moyenne inférieure à 300 nm.

4. Procédé selon l'une quelconque des revendications précédentes, dans lequel on choisit le modificateur de surface dans l'ensemble constitué par l'alcool polyvinylique, un copolymère séquencé tétrafonctionnel résultant de l'addition successive d'oxyde d'éthylène et d'oxyde de propylène à de l'éthylènediamine, la gomme d'acacia, un copolymére séquencé d'oxyde d'éthylène et d'oxyde de propylène, un ester d'acide gras et de polyoxyéthylènesorbitanne et un ester d'acide gras et de sorbitanne.

5. Procédé selon l'une quelconque des revendications précédentes, dans lequel l'agent anticancéreux est le taxol.

6. Procédé selon l'une quelconque des revendications 1 à 4, dans lequel on choisit l'agent anticancéreux dans l'ensemble constitué par le 1,4-dioxyde de 1,2,4-benzotriazine-3-amine, le 1,4-dioxyde de 1,2,4-benzotriazine-7-amine et l'acide rétinoïque.

7. Procédé selon l'une quelconque des revendications 1 à 3, dans lequel l'agent anticancéreux est du 1,4-dioxyde de 1,2,4-benzotriazine-3-amine et le modificateur de surface est de la polyvinylpyrrolidone.

8. Procédé selon l'une quelconque des revendications 1 à 3, dans lequel l'agent anticancéreux est du 1,4-dioxyde de 1,2,4-benzotriazine-7-amine et le modificateur de surface est de la gomme d'acacia.

9. Procédé selon l'une quelconque des revendications 1 à 3, dans lequel l'agent anticancéreux est du 1,4-dioxyde de 1,2,4-benzotriazine-7-amine et ledit modificateur de surface est de la polyvinylpyrrolidone.

10. Procédé selon l'une quelconque des revendications 1 à 3, dans lequel l'agent anticancéreux est du 1,4-dioxyde de 1,2,4-benzotriazine-7-amine et ledit modificateur de surface est un copolymère séquencé tétrafonctionnel résultant de l'addition successive d'oxyde d'éthylène et d'oxyde de propylène à de l'éthylènediamine.

11. Procédé selon l'une quelconque des revendications 1 à 3, dans lequel l'agent anticancéreux est de l'acide rétinoïque et le modificateur de surface est du tyloxapol.

12. Procédé selon l'une quelconque des revendications précédentes, dans lequel la composition est injectable.

13. Procédé selon l'une quelconque des revendications précédentes, dans lequel la composition contient un véhicule pharmaceutique pour celle-ci.

14. Utilisation d'une composition préparée au moyen du procédé selon l'une quelconque des revendications 1 à 13 pour préparer un médicament pour le traitement du cancer.

15. Utilisation selon la revendication 14, dans laquelle le modificateur de surface est présent en une quantité de 0,1 à 90 % en poids par rapport aux particules.

16. Utilisation selon la revendication 14, dans laquelle les nanoparticules ont une dimension moyenne inférieure à 300 nm.

17. Utilisation selon l'une quelconque des revendications 14 à 16, dans laquelle on choisit le modificateur de surface dans l'ensemble constitué par l'alcool polyvinylique, un copolymère séquencé tétrafonctionnel résultant de l'addition successive d'oxyde d'éthylène et d'oxyde de propylène à de l'éthylènediamine, la gomme d'acacia, un copolymère séquencé d'oxyde d'éthylène et d'oxyde de propylène, un ester d'acide gras et de polyoxyéthylène-sorbitanne et un ester d'acide gras et de sorbitanne.

18. Utilisation selon l'une quelconque des revendications 14 à 17, dans laquelle l'agent anticancéreux est le taxol.

19. Utilisation selon l'une quelconque des revendications 14 à 18, dans laquelle la composition est injectable.